

Appl. No. : 09/923,870  
Filed : August 6, 2001

**AMENDMENTS TO THE CLAIMS**

1-48. (Cancelled)

49. (New) A method performed in a computer of producing a genome specific stoichiometric matrix of a microbe, comprising:

determining open reading frames of genes in said microbe;

assigning a function to proteins encoded by said open reading frames by determining the homology of said open reading frames to gene sequences encoding proteins of known function;

determining which of said open reading frames correspond to metabolic genes by determining if the assigned function of said proteins relates to cellular metabolism;

determining substrates, products and stoichiometry for each of said metabolic genes; and

producing a genome specific stoichiometric matrix of said microbe from said substrates, products and stoichiometry.

50. (New) The method of Claim 49, wherein said microbe is *Escherichia coli*.

51. (New) The method of Claim 49, wherein said genes involved in cellular metabolism comprise genes involved in amino acid metabolism, nucleotide metabolism, fatty acid metabolism, lipid metabolism or carbohydrate metabolism.

52. (New) The method of Claim 49, wherein assigning a function comprises performing a homology search using the Basic Local Alignment Search Tool (BLAST).

53. (New) A method of producing an *in silico* representation of a microbe in a computer, comprising:

producing a genome specific stoichiometric matrix of said microbe by the method of Claim 49;

calculating metabolic demands on said microbe;

calculating uptake rates of metabolites of said microbe; and

combining said metabolic demands and said uptake rates with said stoichiometric matrix to produce an *in silico* representation of said microbe.

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54. (New) The method of Claim 53, further comprising performing a flux balance analysis on said *in silico* representation.

55. (New) The method of Claim 53, wherein said metabolic demands on said microbe are calculated by reference to the dry weight composition of said microbe.

56. (New) The method of Claim 53, wherein said uptake rates are calculated by measuring the depletion of substrate from growth media of said microbe.

57. (New) A method performed in a computer for producing a genome specific stoichiometric matrix of a microbe, comprising:

- a) providing the nucleotide sequence of a metabolic gene in the microbe;
- b) identifying the open reading frame of the metabolic gene to determine the corresponding gene product;
- c) assigning a function to the metabolic gene product based on its nucleotide or amino acid homology to other, known metabolic gene products;
- d) determining the substrates, products and stoichiometry for said metabolic gene product based on its assigned function;
- e) repeating steps a) to d) until the substrates, products and stoichiometry of the metabolic genes in said microbe are known; and
- f) producing a genome specific stoichiometric matrix from said substrates, products and stoichiometry of the metabolic genes product in said microbe.

58. (New) The method of Claim 57, wherein the microbe is *Escherichia coli*.

59. (New) The method of Claim 57, wherein said metabolic gene is selected from the group consisting of: genes involved in amino acid metabolism, nucleotide metabolism, fatty acid metabolism, lipid metabolism and carbohydrate metabolism.

60. (New) The method of Claim 57, wherein assigning a function comprises performing a homology search using the Basic Local Alignment Search Tool (BLAST).

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61. (New) A method performed in a computer of producing an *in silico* representation of a microbe, comprising:

producing a genome specific stoichiometric matrix of said microbe by the method of Claim 57;

calculating metabolic demands on said microbe;

calculating uptake rates of metabolites of said microbe; and

combining said metabolic demands and said uptake rates with said stoichiometric matrix to produce an *in silico* representation of said microbe.

62. (New) The method of Claim 61, wherein said microbe is *Escherichia coli*.

63. (New) The method of Claim 61, wherein said metabolic demands on said microbe are calculated by reference to the dry weight composition of said microbe.

64. (New) The method of Claim 61, wherein said uptake rates are calculated by measuring the depletion of substrate from growth media of said microbe.

65. (New) The method of Claim 61, further comprising performing a flux balance analysis on said *in silico* representation of said microbe.